

CLAIMS

1. A method for producing a template of sequences of chemical or biological molecules formed from different chains of molecules M₁, M₂,.....M_n, by in situ synthesis on a structured substrate with 5 microcuvettes, comprising the following steps:

1) functionalization of the microcuvettes of the substrate by functional groups capable of forming a covalent bond with the molecules M₁, M₂,.....M_n;

10 2) deposit of a protection polymer on at least one of the microcuvettes by microdeposition of drops from said polymer to form caps of solid polymer on the selected microcuvette(s);

15 3) carrying out one or more chemical reactions to provide coupling of a first molecule M₁ on the functional groups of the microcuvettes not covered with protection polymer;

20 4) removal of the protection polymer on the microcuvettes covered with this polymer, after the first reaction or one of the following reactions carried out in step 3); and

25 5) again carrying out steps 2), 3) and 4) to obtain desired chaining sequences on each of the functionalized microcuvettes.

2. The method according to claim 1, wherein step 1) consists of:

a) functionalizing the total surface of the substrate and microcuvettes with the functional groups;

- b) depositing a protection polymer on all the microcuvettes by microdeposition of drops from said polymer to form caps of solid polymer on all the microcuvettes;
- 5 c) reacting the functional groups present on the substrate around microcuvettes with a nonlabile blocking group under the conditions used for the chemical coupling reactions and for the removal of the protection polymer; and
- 10 d) removing the protection polymer from all the microcuvettes.
3. The method according to claim 2 wherein after step a), the functional groups are protected by a labile protective group and deprotection of the 15 functional groups is carried out after step b) and before carrying out step c).
4. The method according to any of claims 1 to 3 wherein functional groups of the microcuvettes are protected by a labile protective group and step 3) 20 comprises a first deprotection reaction of the functional groups.
5. The method according to claim 3 or 4 wherein the protective group is a trityl or trityl-derived group.
- 25 6. The method according to any of claims 1 to 5 wherein the sequences of molecules are oligonucleotides.
7. The method according to claims 5 and 6 wherein step 3) is a phosphoramidite synthesis cycle of 30 oligonucleotides which consists of removing the trityl

groups, of coupling with the nucleotide, reacting the functional groups, which have not been coupled with the nucleotide, with a blocking group and oxidizing the phosphoramidite group of the nucleotide into a 5 phosphate.

8. The method according to claim 7 wherein step 4) is carried out with removal of the protection polymer after coupling of the nucleotide.

9. The method according to claim 8 wherein the 10 protection polymer is selected from the group formed by polymers and their derivatives of polyvinyl alcohols, polystyrenes, polyvinyl carbazoles and polyimides.

10. The method according to claim 7 wherein step 4) is carried out for removing the protection polymer, 15 after step 3) for removing the trityl groups.

11. The method according to claim 10 wherein the protection polymer is polyhydroxystyrene and detritylation is carried out in dichloromethane.

12. The method according to claim 10 wherein the 20 protection polymer is polystyrene or polyvinyl carbazole, and detritylation is carried out in acetonitrile.

13. The method according to claim 10 wherein the protection polymer is polyethylene oxide and 25 detritylation is carried out in toluene.

14. The method according to claim 10 wherein the protection polymer is polyvinyl alcohol and detritylation is carried out in dichloromethane.

15. The method according to any one of claims 10 30 to 14 wherein step 4) for removal of the protection

polymer is carried out by rinsing by means of a solvent of the polymer by adding this rinsing step in the cycle of an automatic synthesizer of oligonucleotides.

16. The method according to any one of claims 1
5 and 2 wherein the protection polymer is deposited by means of a microdeposition robot by the ink jet printing technique or dispenser of calibrated microdrops.

17. The method according to claim 1 or 2 wherein,
10 as the polymer is deposited from a solution in a solvent, the polymer film formed is annealed by heating the substrate to a temperature of 50 to 100°C before carrying out step 3) or step c).